

1/9

**CE Conditions**

Capillary Electrophoresis: TSP CE1000  
 Capillary (type/length): Bare fused  
 silica(375 $\mu$ m O.D., 50 $\mu$ m I.D.yeffective  
 32cm. total 40cm.

Applied Voltage: 25kV

Injection: Hydrodynamic, 5sec.

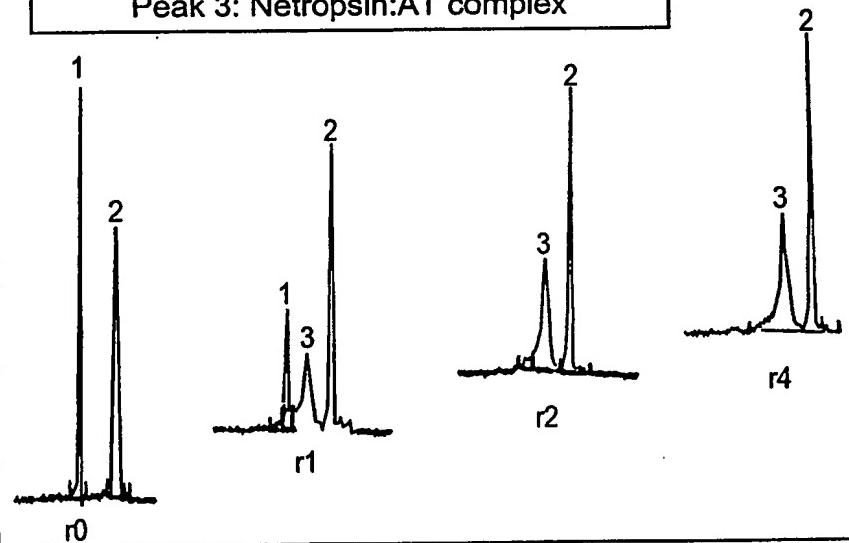
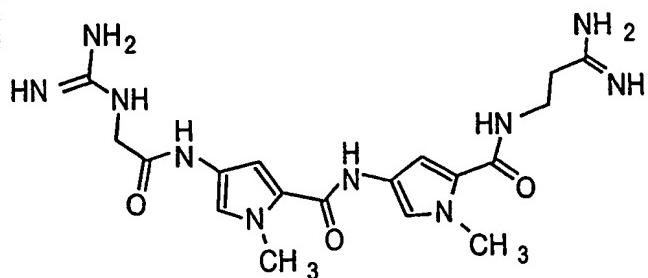
Capillary temperature 20°C  $\pm$ 0.1°C

Buffer: 0.02M Borate buffer, pH7.5

UV detection: 260nm.

DNA Cone: 20 $\mu$ M

**Key:**  
 Peak 1: AAATTATATTAT (AT)  
 Peak 2: GGGCCGCGCCGC (GC)  
 Peak 3: Netropsin:AT complex

**Netropsin**

Competitive binding studies with netropsin.

**Fig. 1**

2/9

**CE Conditions**

Capillary Electrophoresis: Agilent G1600

Capillary (type/length): Bare fused  
silica(375 $\mu$ m O.D., 50 $\mu$ m I.D. effective  
25cm. total 33.5cm.

Applied Voltage: 25kV

Injection: Pressure, (50mBar,5sec)

Capillary temperature 20°C  $\pm$ 0.1°C

Buffer: 0.02M Borate buffer, pH7.5

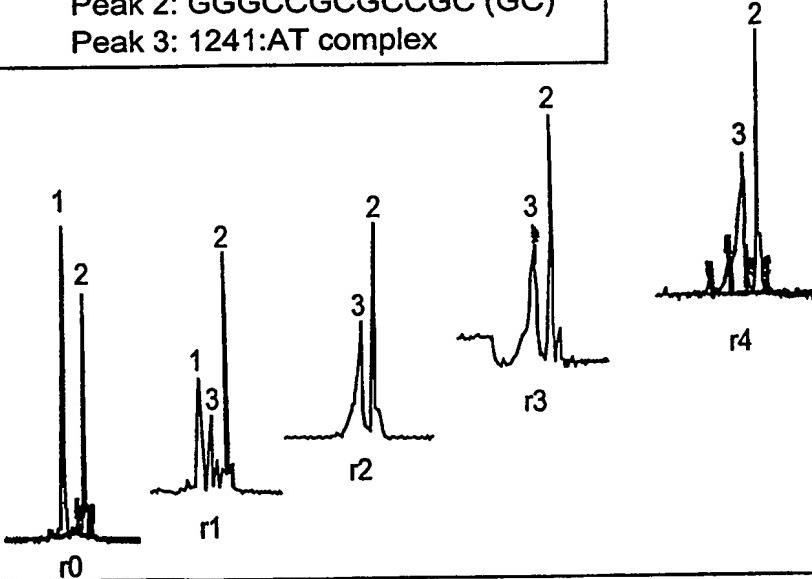
UV detection: 260nm.

DNA Cone: 20 $\mu$ M

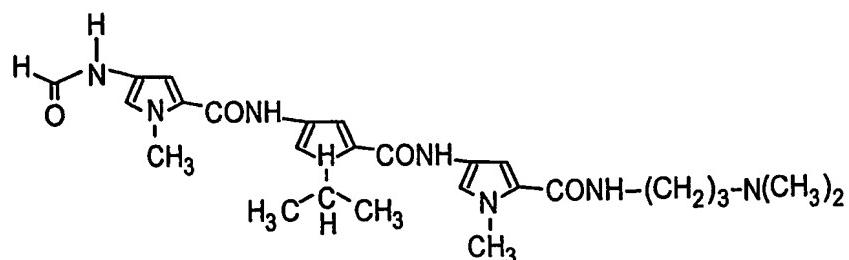
Key: Peak 1: AAATTATATTAT (AT)

Peak 2: GGGCCGCGCCGC (GC)

Peak 3: 1241:AT complex



12/41



Competitive binding studies with compound 12/41

***Fig.2***

3/9

CE Conditions

Capillary Electrophoresis: Beckman P/ACE MDQ.

Capillary (type/length): Bare fused SILICA(375  $\mu$ m O.D., 50  $\mu$ m I.D. effective 21cm, total 31.2 cm.

Applied Voltage: 25kV

Injection: Pressure, (20 P.S.I., 0.5 sec)

Capillary temperature 20°C  $\pm$  0.1°C

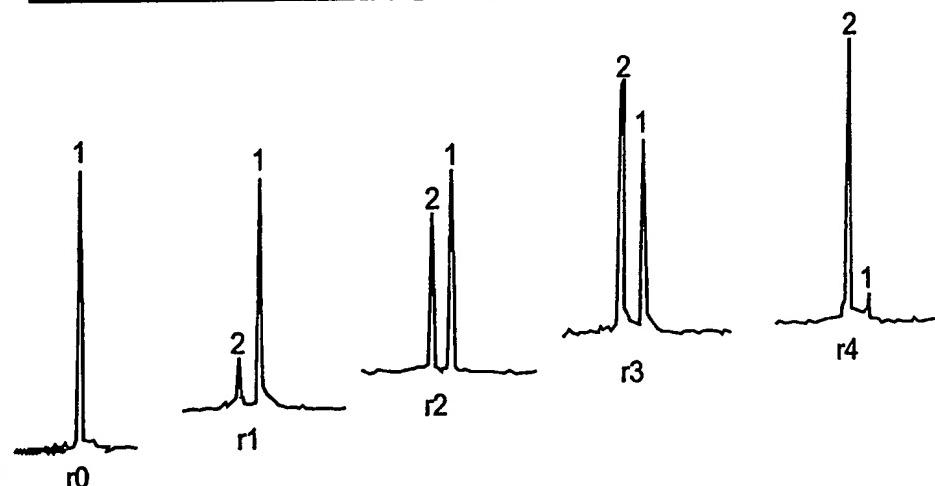
Buffer: 0.02M Borate buffer, pH7.5

UV detection: 260nm.

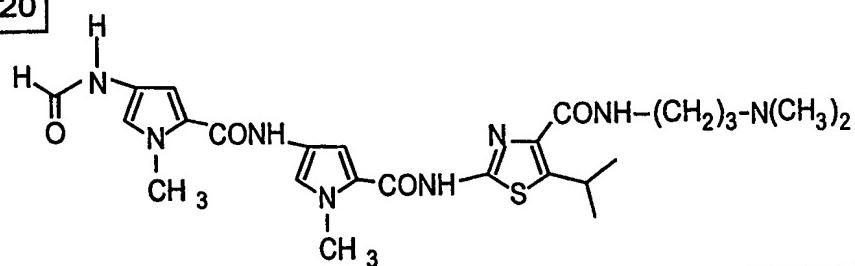
DNA Cone: 20  $\mu$ M

Key: Peak 1: CGACTAGTCG (ACTA)

Peak 2: 1320:ACTA complex



13/20



Capillary electrophoresis studies of the interaction between compound 13/20 and DNA decamer CGACTAGTCG.

**Fig.3**

4/9

CE Conditions

Capillary Electrophoresis: Beckman P/ACE MDQ.

Capillary (type/length): Bare fused SILICA(375 µm O.D., 50 µm I.D.yeffective 21cm, total 31.2 cm).

Applied Voltage: 25kV

Injection: Pressure, (20 P.S.I., 0.5 sec)

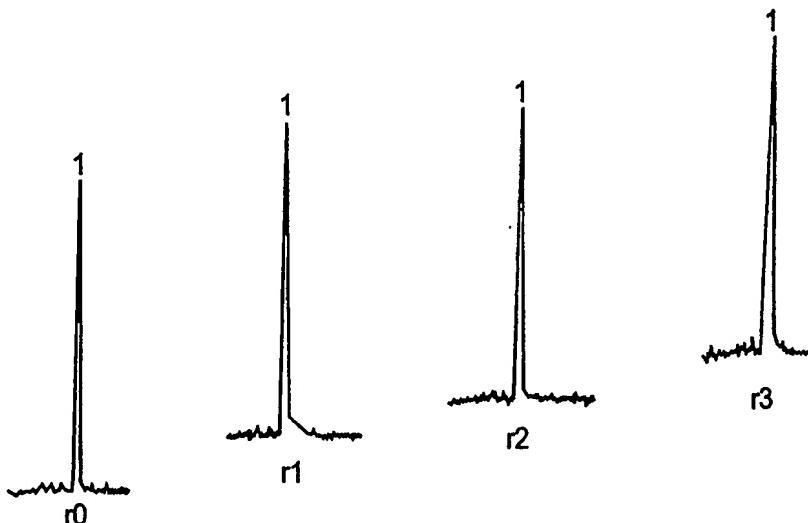
Capillary temperature 20°C ±0.1°C

Buffer: 0.02M Borate buffer, pH7.5

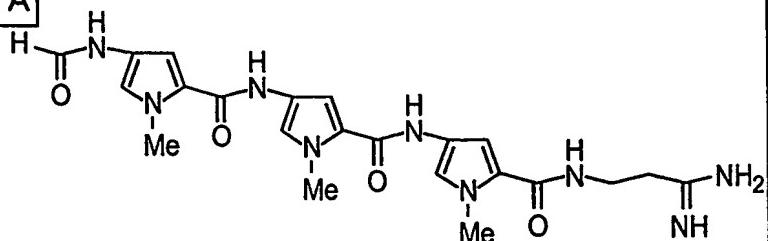
UV detection: 260nm.

DNA Cone: 20µM

Key: Peak 1: CGACTAGTCG (ACTA)



Distamycin A



Capillary electrophoresis studies of the interaction between distamycin and DNA decamer CGACTAGTCG.

**Fig.4**

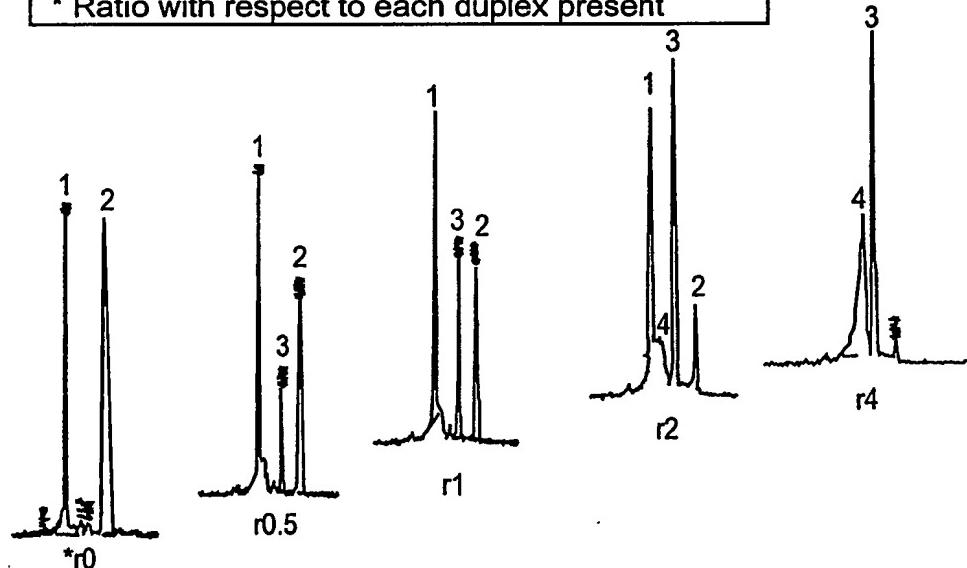
5/9

CE Conditions

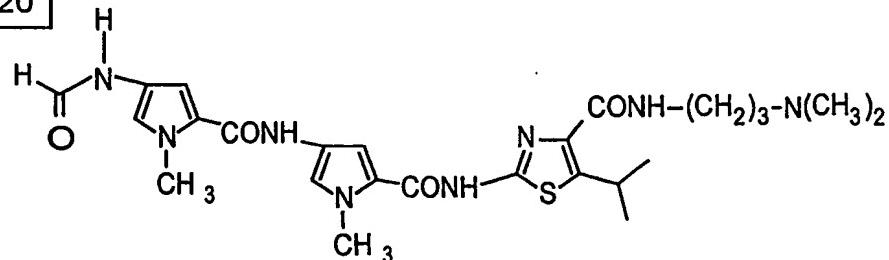
Capillary Electrophoresis: Agilent G1600  
 Capillary (type/length): Bare fused SILICA(375 µm O.D., 50 µm I.D. effective 25cm, total 33.5 cm.  
 Applied Voltage: 25kV  
 Injection: Pressure, (50mBar, 5 sec)  
 Capillary temperature 20°C ±0.1°C  
 Buffer: 0.02M Borate buffer, pH7.5  
 UV detection: 260nm.  
 DNA Cone: 20µM

Key: Peak 1: AAATTATATTAT (AT).  
 Peak 2: CGACTAGTCG (ACTA).  
 Peak 3: 1320: ACTA complex.  
 Peak 4: 1320: AT complex.

\* Ratio with respect to each duplex present



13/20



Competitive binding studies with compound 13/20

*Fig 5*

6/9

**CE Conditions**

Capillary Electrophoresis: Beckman P/ACE  
MDQ.

Capillary (type/length): Bare fused Silica(375 $\mu$ m O.D., 50 $\mu$ m I.D.yeffective 21cm, total 31.2 cm.

Applied Voltage: 25kV

Injection: Pressure, (20 P.S.I., 0.5 sec)

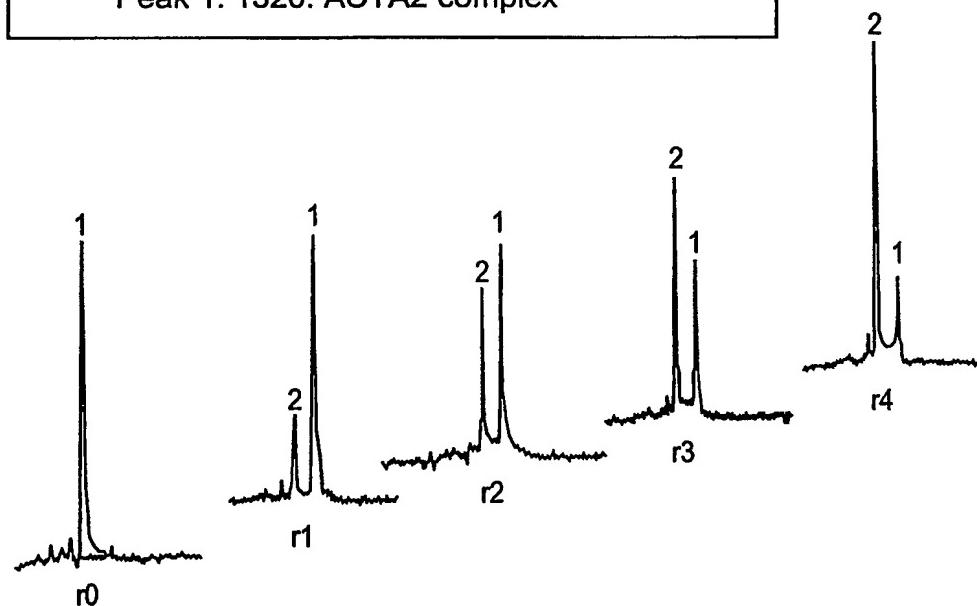
Capillary temperature 20°C  $\pm$ 0.1°C

Buffer: 0.02M Borate buffer, pH7.5

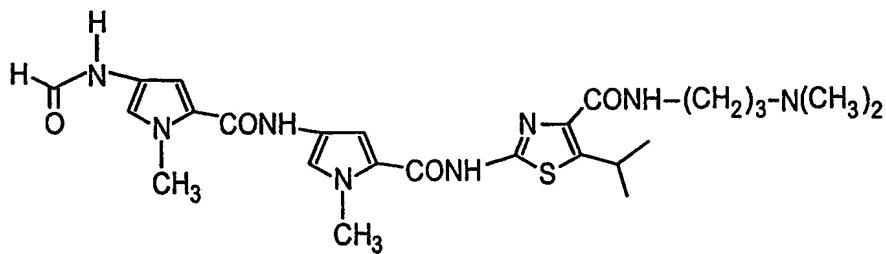
UV detection: 260nm.

DNA Cone: 20 $\mu$ M

**Key:** Peak 1: CGACTAGTGG (ACTA2)  
Peak 1: 1320: ACTA2 complex



1320

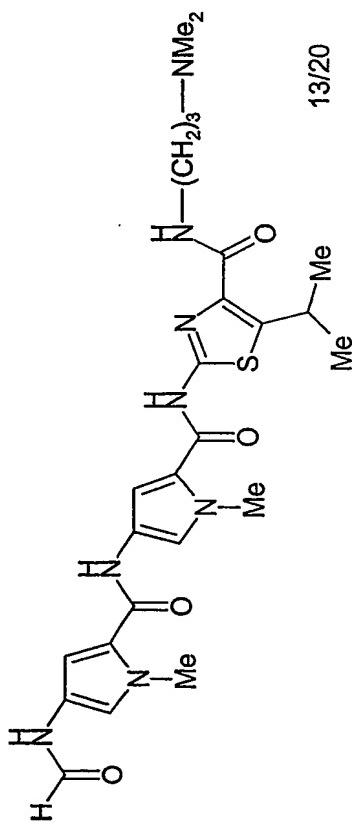


Capillary electrophoresis studies of the interaction between compound 13/20 and DNA decamer CGACTAGTGG

**Fig 6**

7/9

## Footprinting studies with compound 13/20

**Fig. 7**

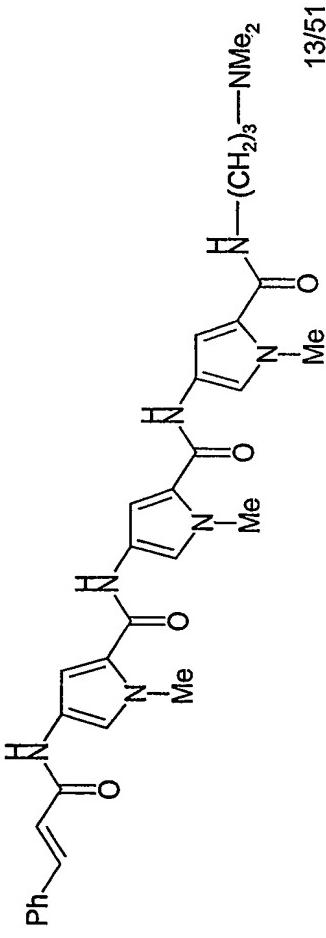
5'-GGATCCATATGCCGAAATACACATGGCCAACTGCACCTAGTCGTAGCGCGATCAAGGTAAAGCTCCCGTTCTATCC  
x XXXXXXXXXX

TGGTATAAGCAARTAGGGCGTGAAGAGTTATGTAAAGTACCGTCCGGTGGGGCTCTGTTTGTCATCTCAGCCTCGAATGCGGATCTC

(Experiment carried out at 0.03  $\mu$ M of compound 13/20)

8/9

## Footprinting studies with compound 13/51



13/51

Fig. 8

5'-GGATCCATATGGGGCAATAACACATGGCCAACTGGCAACTAGTCGTAGCGGATCAAAGGTTAACGGTTAAGCTCCGGTCTATCC  
xxxxx(0.1)

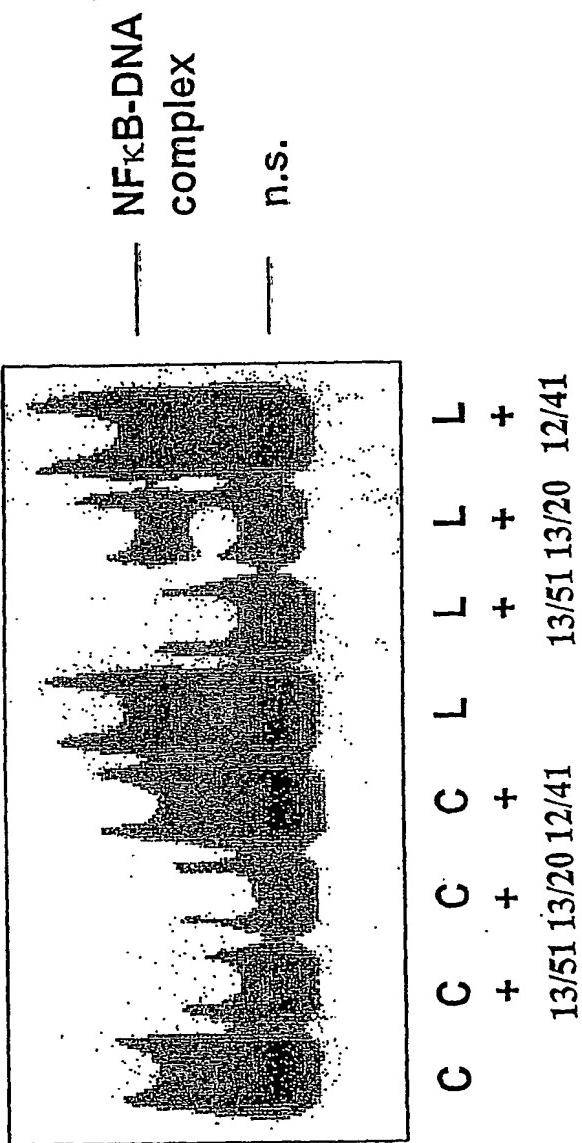
TGGTATAAGCAATTAGGGCGTGAAGAGTTATGTAAGGTACGTCCGGTGGGTCTGTTGTCATCTCAGGCCATCGAATGGGGATC  
xxxx(3) xxxxx(0.1)xxxx(1)xxxx(0.01)  
xxxxx(0.1)

The numbers in brackets refer to the concentration of compound 13/51 (in  $\mu\text{M}$ ) at which the relevant footprint was observed.

9/9

Effect of DNA Minor Groove Binding Compounds upon LPS-  
Stimulated NF $\kappa$ B-DNA Binding Activity in Murine Macrophages

lanes: 1 2 3 4 5 6 7 8



C = control; n.s. = non-specific

Fig. 9